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Growth of Black Pine in Cultures under Influence of some Pathogens in the Area of Šumadija, Serbia

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ABSTRACT

This study dealt with growth culture conditions of the black pine in Sumadija, Serbia. It aimed to determine the effect of the pathogenic fungus, *Fomes annosus* in healthy and infected cultures of black pine and its impact in its growth. Research was conducted in two experimental fields and in two series. Effect of the fungus was followed at different soil depths and root zones of pine as well in old and young crops. The tests were carried out on nutritive substrates and land suspensions.

Key words: Black pine, Culture, Soil, *Fomes annosus,* Serbia.

INTRODUCTION

The land is very dynamic environment where turbulent biochemical processes continually happen, conditioned by the huge potential of soil microorganisms that make the process of decomposition of forest litter, humus synthesis and its mineralization. While making a study, each biocenosis was characterized by a certain microbiocenosis (Bakys *at al.* 2006) and that in silvibiocenois fungi of the genus *Fomes*. A certain place is occupied as decomposers of ligneous materials (Whitney and Irwin, 2005) which is due to the very complex chemical composition difficult to decompose. Considering qualitative composition of microorganisms in silvibiocenosis conditioned by nature of the organic matter in forest litter and its population of microecological environmental conditions the change of these factors may completely change the relationship among individual members of microbiocenosis. Silvicultural measures (cleaning and thinning) change the microecological conditions in silvibiocenosis, ingestion of other species and inadequate fertilization, biological balance even moves more, which might affect the populations of soil microorganisms.

Considering all of these, the task for studying soil microflora was set including its dynamics and relationships and value of the role played by the soil fungi *Fomes annosus* among different groups of microorganisms under different circumstances.

MATERIALS AND METHODS

On the experimental fields in the forests and plantations of *Pinus nigra* in central region of Serbia, tests were conducted on perennial populations of microflora in soil and rhizosphere in healthy and infected stands in which the pathogenic fungus *F. annosus* expressed varying intensities. Land and rhizosphere microflora was tested on agar nutrient seeding soil by applying soil dilution of 1: 1000. Land agar was used for the total number of microorganisms (Dahlberg *et al.*, 2001) for fungi and the agar synthetic agrochemicals Krasiljnikov actinomycetes and Edgie's agar was used for aligotrophic microorganisms. In the rhizosphere microflora, two zones were investigated, namely the land that clings to the hair roots and rhizosphere of root veins. The research area is forest management area Kragujevac. This area consists of forestry unit in Gornji Milanovac and one in Kragujevac as well. Concrete measures of black pine cultures were done in the area of Gruža-Lepenica-Jasenica forest, which belongs to forestry unit of Kragujevac. Gruža-Lepenica's forest is spread in the area of 2950 ha. 285 ha are forest cultures. That's almost 10 percent. It should be mentioned that these are mostly black pine cultures.

RESULTS AND DISCUSSION

Soil microflora in the stands of black pine in central region of Serbia (I Series)

As this is a land of light mechanical composition-brown loam, with a lot of low humus content (Lygis et *all*,2004) the total number of microorganisms in soil was very high in the surface horizon of up to 13 cm depth (Fig. 1) regardless of the degree of contamination of the stand. When depth increase up to 40 cm, total number of microorganisms rapidly decreased. At greater depths, total number of microorganisms rapidly decreased, up to 40 cm depth was still variable. At greater depth of microorganisms, it became very low. In healthy stands and the stands of the first degree of infestation, total number of microorganisms was uniform throughout the profile, without rapid transitions. In the stands of II and III degree of infection, the total number of microorganisms slightly increased in the surface horizons to 3 cm, while with increasing depth, it rapidly decreased with some fluctuation in the horizon of 13-40 cm. (Fig . 1).

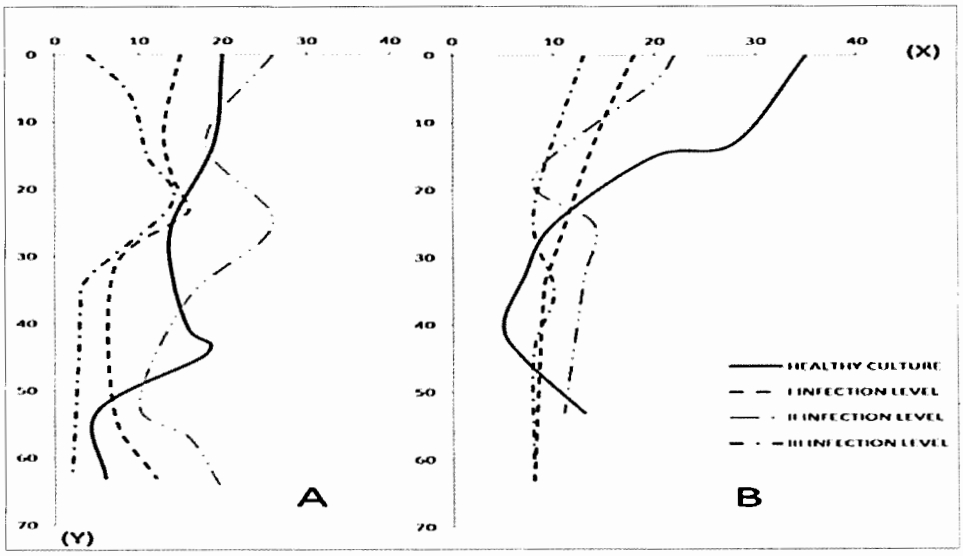

Fig. (1): Movement of actinomycetes (A) and mushroom (B) in the cultures of black pine in the areacentral region of Serbia, (33 years old culture). Depth land, in cm (Y), Number of microorganisms in 000-(X).

In the stands of II and III degree of infestation, total number of amonificational microorganisms, which depends on the amount and state of the organic matter in the soil, was considerably bigger up to 3 cm of the depth, and rapidly decreases with the depth. In healthy soil under stands, the degree of infection and number of microorganisms were less amonifictional but uniform up to 40 cm depth. Such uniform number of amonificational organisms, depth profiles in health contents indicated that organic matter is distributed evenly and that there is no delay in its decomposition. The ppopulation of oligotrophic microorganisms was significantly higher in soil infected stands, while in the soil healthy stands free from these microorganisms , the number was extremely low and coincided with the number of amonificational microorganisms. In healthy stands of soil, the population of fungi was extremely high in the horizon of up to 13 cm depth. At greater depths, the population of these microorganisms was decreasing and was equated with the number of these microorganisms in soil infected stands, which is a relatively low profile over the entire length (Fig. 1).

Soil microflora in the stands of black pine in central region of Serbia (II series):

The stands of black pine varied in degrees of infection at I, II, III, in which experimental areas were set and developed on the brown-podzolic soils. These soils are deep, of light mechanical composition, with high content percentage of coarse sand, with a strong pronounced acidity and a high percentage of humus to a depth of 20-30 cm (DeBano, 2000).

According to the microbiological analysis of land in the stands, it showed a very low biogeny, and differences in total numbers of microorganisms between healthy and infected stands were minimal (Fig. 2). The number of amonificational microorganisms was very low. The differences in numbers of the amonificational microorganisms according to the degree of infection were minimal. In healthy soils and those with low number of infected stands, population of oligonitrophilous microorganisms was very low, but it was extremely high in soils with heavily infected content, since in the horizon, it's located up to 20 cm depth, while the number of oligonitrophilous microorganisms reached 8 to 9 millions. So high population of microorganisms indicated that in infected stands of black pine, limited nitrogen nutrition becomes even more pronounced rather than known in the stands of white pine.

Population of actinomicity was markedly higher than in the soil with healthy and poorly infected stands (Fig. 2) because they were registered severely in infected stands to a depth up to 20 cm and in a minimum number. Significantly higher number of actinomycetes in healthy and with poorly expressed stands indicating complete decomposition of organic matter in the soil, because it is known that actinomycetes participating in decomposition of organic matter, which certainly affects the quality of humus and the content of available elements of nutrition. The number of fungi was also significantly higher in the soil of healthy and poor infected stands of the entire length of the profile. In heavily infected stands, fungi were recorded only in the horizon up to 10 cm depth (Fig. 2).

The presented data on the number of microorganisms suggest that the overall biological value under black pine stands in Šumadija is relatively low. Difference in population between healthy soil of microorganisms and infected stands were reflected in a large population of amonificational organisms in soil infected stands, which points to that the land is organogenic and that

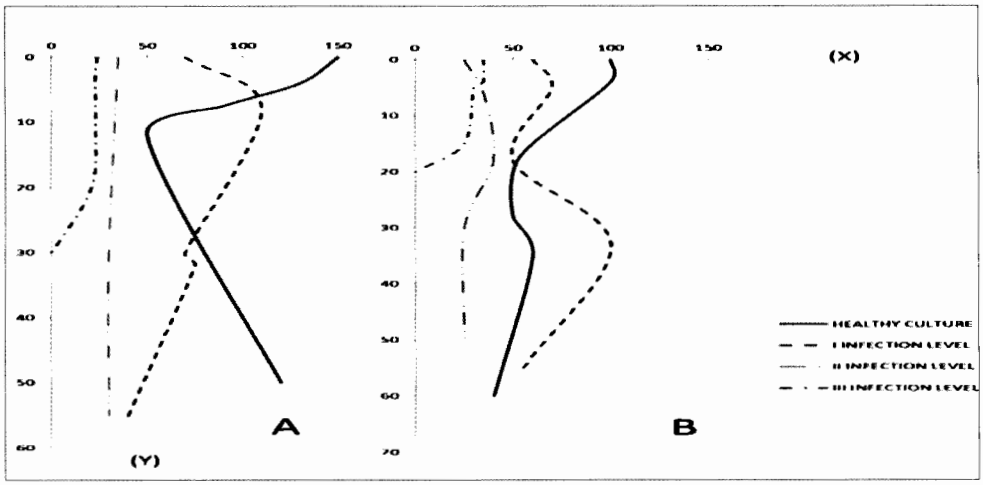

Fig. (2): Movement of actinomycetes (A) and mushroom (B) in the cultures of black pine in the area of central region of Serbia, (55 years old culture).Depth land, in cm (Y), Number of microorganisms in 000-(X).

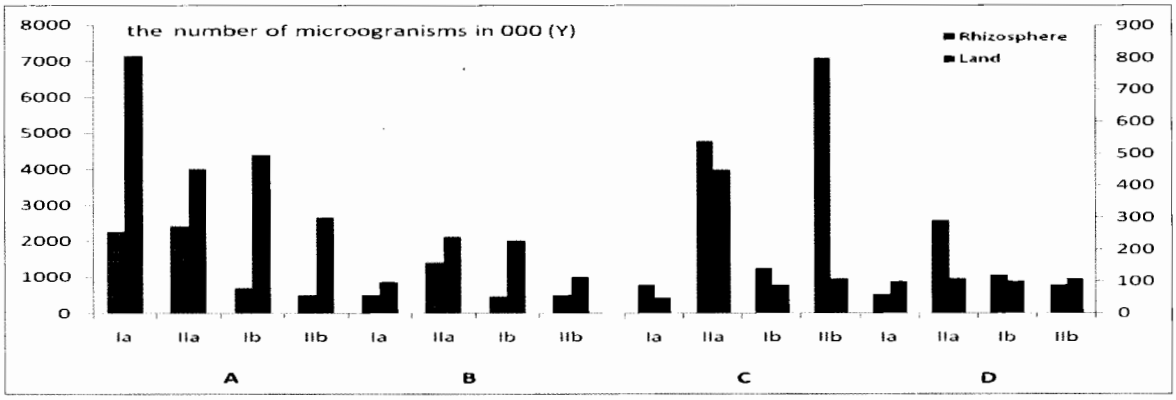

Fig. (3): Movements in the number of microorganisms in the rhizosphere, in the culture of the black pine-under crops in the area -central region of Serbia

-(A) total number of microorganisms; **(B)** amonificational microoorganims; **(C)** actinomycets; **(D)** fungi

I - infected trunk; **II** - healthy trunk; **a** - a trunk within; **b** - a trunk in remote area.

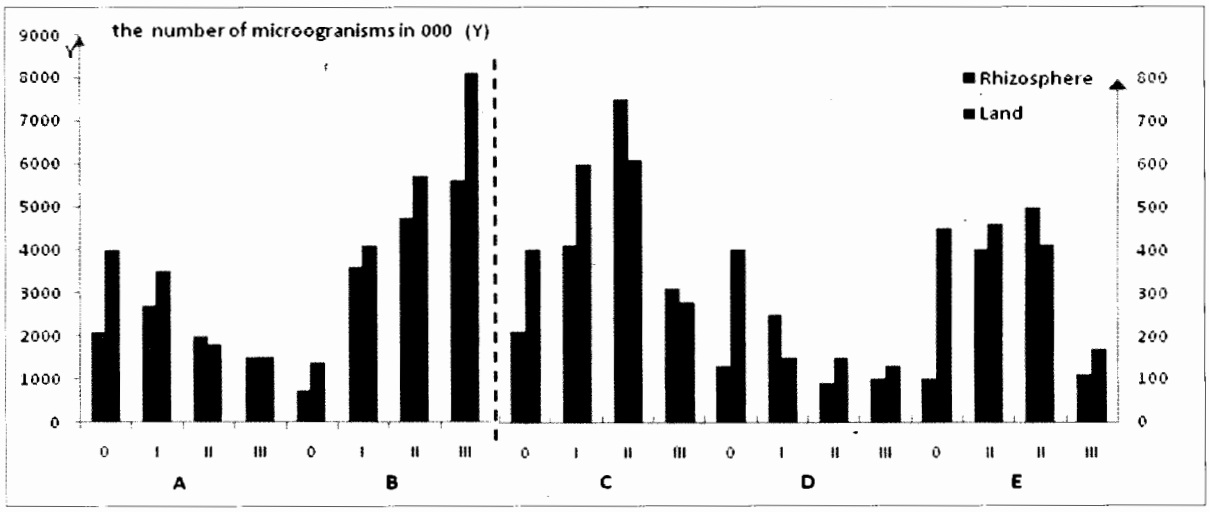

Fig. (4): Movements in the number of microorganisms in the rhizosphere, in the culture of the black pine-older cultures in the area - central region of Serbia-(Y—the number of microorganisms in 000).

(A) total number of microorganisms; **(B)** oligonitrofile microorganisms; **(C)** amonificational; **(D)** actinomicetes; **(E)** fungi

0 - healthy trunk; **I** - first degree on infection; **II** - the second degree of infection; **III** - degree of infection.

a large proportion of nutrients, blocked in organic form. Much smaller number of fungi and actinomycetes contributes to it, particularly involved in decomposition of mineralization of organic matter and humus. High population of oligonitrophilous microorganisms in soil infected stands, indicated unfavorable nitrogen regime, because it is known that these microorganisms develop in the soil with low available nitrogen, since they are able to take ammonification under adverse conditions (Akkemik, 2001) and in the toughest conditions for nitric nutrition to compensate this element by nitrification (Cairney and Bastias, 2007). Low nitrogen content in podzolic soils has not indicated yet (Menkis, 2006)

Soil and rhizosphere of infected stands, where the nitrogen regime even the number of microorganisms, the antagonist was much smaller which allowed a greater population of the fungus, *F. annosus*. This conclusion was proved by (Ludovici and Kress, 2006) since the introduction of nitrogen fertilizers in the soil with the altered conditions of nitrogen nutrition increased an antibiotic activity of microorganisms antagobista to the fungus *F. annosus.*

Finally, it could be concluded that according to the results of the present study, the regime and nitrogen nutrition extremely should be given a special attention to high population of oligonitrophilous microorganisms in soil infested stands.

Analysis of the rhizosphere microflora in infected and healthy stands

Rhizosphere is the most active and dynamic part of the land in which complicated interrelationships of plants and microorganisms occur. Big differences in numbers of the individual groups and rhizosphere microorganism of investigated trunks showed that under the influence of root and total number of microorganisms increased significantly. Still a stronger effect is in healthy trees, especially when it comes to fungi and actinomycetes. The data (Figs. 3 and 4) illustrate that the numbers of fungi and actinomycetes were significantly higher in healthy trees, whether they are within or remote. These data suggest that interaction of healthy trees roots and soil microorganisms are active and result in a very high population of microorganisms increasing resistance to disease. Effect of oligonitophilic microorganisms in the rhisosphere of infected trees was very high, where the number of these organisms reached about 9 million Fig. (4). This indicates that in the sphere of the narrowest roots of infected trees, nitrogen regime was exacerbated, which certainly has the consequences on nitrogen nutrition of plants.

It can be concluded that the effect of the roots of healthy trees of black pine is very high, and it is reflected in the increased population of total microflora, fungi and actinomycetes in the area of the root. Only the number of oligonitrophilous of microorganisms significantly increased in the sphere of the roots of infected trees. Qualitative analysis of the population of some groups of microorganisms in the soil and rhizosphere pointed to the outstanding differences in the composition of the population and land microflora on the infected and uninfected areas. The differences are as follows: population of fungal microflora and actinomycetes was significantly higher in soil and rhizosphere of healthy stands, while the number of oligonitrophilous microorganisms was crucially higher in infected stands. The difference in total numbers of the microorganisms and amonificational microorganisms was very small. Differences were significant in the qualitative composition of microflora and actinomycetes. All this indicates that the soil infected and healthy stands differed in biogeny and that among other things in the area, the causes of the expansion of the fungus *F. annosus* should be searched. Lots of healthy and infected stands with the fungus, differ in population and composition of soil microflora and among other things also the explanation for expansion of the fungus. The total biogenic of the soil under the stands of black pine was averaging, if different habitats were included, average or relatively turned low, but only if it was viewed through some average values. In the soil infected stands, a slightly greater number of amonificational and oligonitophilic microorganisms were present in a large population, since the fungi and actinomycetes occur in small numbers.

In the land of healthy stands, total number of microorganisms was greater than amonificational and oligonitophilic microorganisms with low population, yet fungi and actinomycetes were present in large numbers. In the qualitative composition of soil microflora, no outstanding difference between infected and healthy stands was found. Rizosphere effect of healthy black pine trees was very high and was reflected in the increased population of total microflora of fungi and actinomycetes in the area of the root. The chart presents the growth and increment of black pine in cultures, influenced by microflora and all the factors mentioned.

It is notable that the growth began to decline, as the impact of decaying (*F. annosus*) of Pine cultures vulnerability of rot *F. annosus* was great. Dynamics of drying is 2% per year. If the current dynamics continue for 50 to 60 years, the current inventory of pine cultures would disappear. That is why it is urgent to take measures concerning care and sanitation (Fig. 5).

In conclusion, the investigated territory in central Serbia at the analyzed cultures of black pine, can be

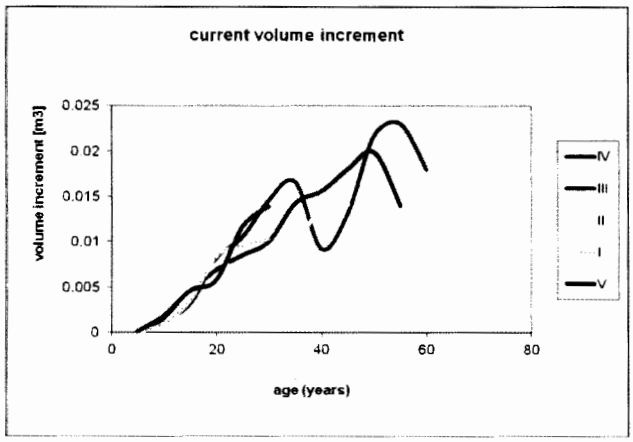

Fig. (5): Increment volume for cultures of black pine.

concluded as:

- Healthy and infected plot stands with mushroom "*F. annosus*" differ in population and composition of soil microflora.
- -Total biological value in-younger-infected-investigated stand soil culture (33 years old) was very low.
- From lands–older-infected-investigated culture (55 years), a slight higher number of amonification microorganisms, compared to younger infected culture-and oligonitophilic microorganisms, compared to younger culture, occurred in vast populations. Fungi and actinomycetes were represented in slightly greater numbers than in the younger stand culture.

It is worthy to note that in healthy stands of 33 years age, total number of microorganisms was greater than amonification and oligonitophilic microorganisms. Mushrooms and actinomycetes were represented in higher numbers. At healthy stands of 55 years age, total number of microorganisms was greater ,than amonifikacion and oligonitophilic microorganisms and showed a slightly higher population than younger Hi culture.

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